

Figure 1

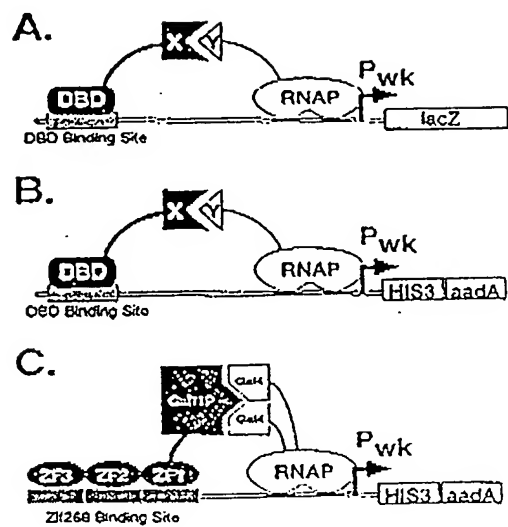


Figure 2

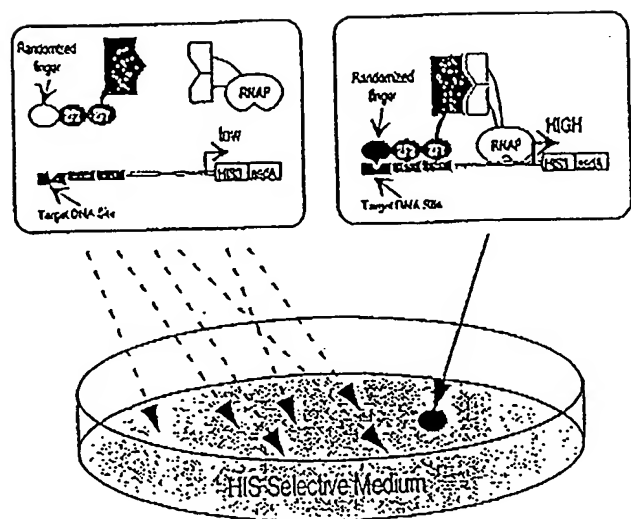


Figure 3

A. TATA Subsite Selection:

3' AAA 5' 3' AAA 5'

Phage Display
Consensus:

Group I:

-1	1	2	3	5	6
Q	+	-	N	-	-
Q	R	G	N	L	V
Q	K	T	N	M	V
Q	K	Y	N	I	L
Q	R	Y	N	V	V
Q	K	C	N	M	V
Q	K	C	N	H	V
Q	R	C	N	M	V
Q	R	C	N	K	T
Q	L	G	N	M	V
Q	K	G	N	K	V
Q	L	G	N	K	V

(3)

-1	1	2	3	5	6
N	S	G	A	-	N
N	S	G	A	Y	N
N	S	G	A	W	N
N	S	G	A	F	N
N	S	G	A	Y	N
N	S	G	A	Y	N
G	S	G	A	Y	N

(4)

Group II:

Q	R	G	N	K	K
Q	R	C	N	K	K
Q	R	G	N	V	K
Q	K	G	N	K	K
Q	K	G	N	Y	K
Q	K	H	N	K	K
Q	K	G	N	S	R
Q	S	G	N	K	K

(2)

Q	K	S	N	L	R
Q	K	A	N	T	K
Q	K	A	N	K	L
Q	L	S	N	K	K
Q	P	G	N	K	K
Q	K	G	N	M	V
Q	K	S	N	M	V
Q	K	H	N	L	R

(2)

C. NRE Subsite Selection:

3'ACT5'

↑ ↑ ↑

-1	1	2	3	5	6
N	S	G	S	W	K
N	S	G	S	W	K
N	H	G	S	W	K
N	S	G	S	H	K

(15)
(2)

Group I:
first selection:
second selection:

Phage Display
Consensus:

T - T N - S

B. p53 Subsite Selection:

	3'TGT5'							-1 1 2 3 5 6						
	-1	1	2	3	5	6		-1	1	2	3	5	6	
<u>Phage Display</u>														
<u>Consensus:</u>	H	-	H	H	-	-								
<u>Group I:</u>	H	K	H	H	K	A	(2)							
	H	L	H	H	K	A								
	Q	R	W	L	M	R		W	R	S	S	L	V*	
	Q	R	W	L	R	R		W	Q	S	S	K	V*	
	Q	D	N	L	R	R		W	Y	S	S	I	V*	
	Y	K	R	D	N	R		F	D	S	S	R	A*	
<u>Group II:</u>	A	R	W	L	N	R		E	R	W	R	N	R	(4)
	E	R	W	R	N	R		E	R	W	R	N	R	
	E	R	W	R	R	R		E	R	W	R	I	R	
	E	R	W	R	R	R		E	R	W	R	S	R	
	E	R	W	R	R	R	(2)	E	R	W	R	V	R	
	E	R	W	R	R	R		E	R	W	R	V	R	
	E	R	W	R	R	R		E	R	W	R	V	R	
	E	K	W	R	R	R	(2)	E	R	W	R	L	R	
	E	G	W	R	R	R		E	K	W	R	T	R	
	E	R	W	R	K	R	(2)	E	N	W	R	K	R	
	E	R	W	R	K	R		E	Q	W	R	K	R	
	E	R	W	R	K	R		E	R	W	R	M	R	

D. Group III Fingers:

	-1	1	2	3	5	6		-1	1	2	3	5	6
From the													
<u>TATA Selection:</u>	R	R	W	L	K	L		R	K	W	L	Q	L
	R	K	W	L	K	L		R	K	W	L	R	L
From the													
<u>p53 Selection:</u>	R	K	W	L	R	L		R	K	W	L	M	L
	R	K	W	L	R	L		R	K	W	I	N	L
	R	K	W	L	R	L		R	K	W	Y	Q	L
	R	K	W	L	R	L							
From the													
<u>NRE Selection:</u>	R	K	W	L	R	L		R	A	W	L	K	L
	R	K	W	L	K	L		R	K	W	Y	R	L
	R	K	W	L	K	L							

Behavior of different Fluorescent Proteins in the Bacterial Two-Hybrid System

Fig. 4

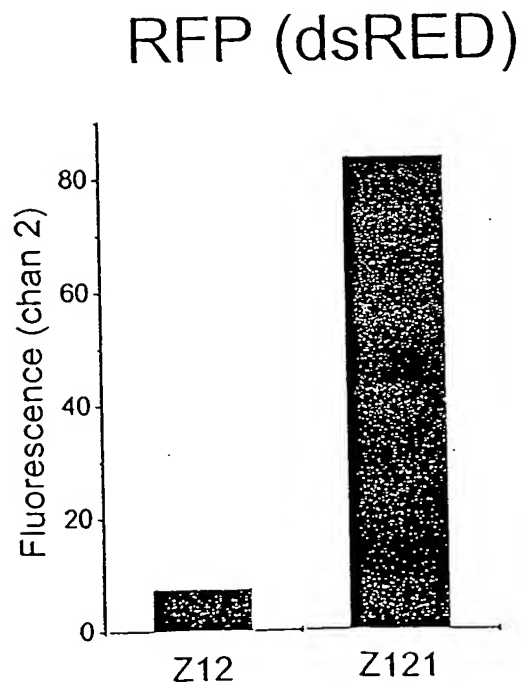
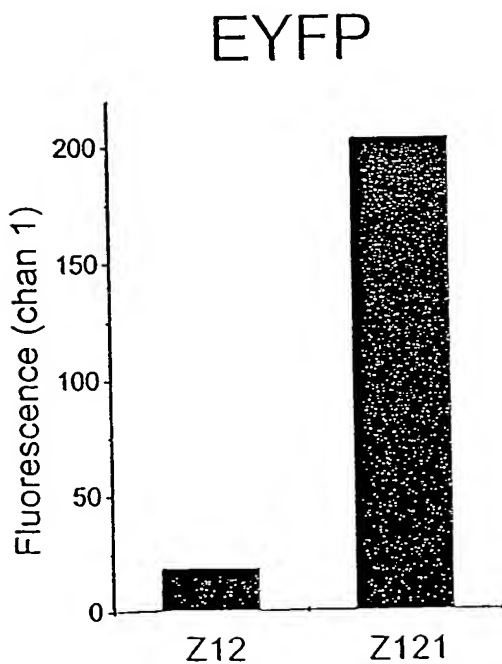
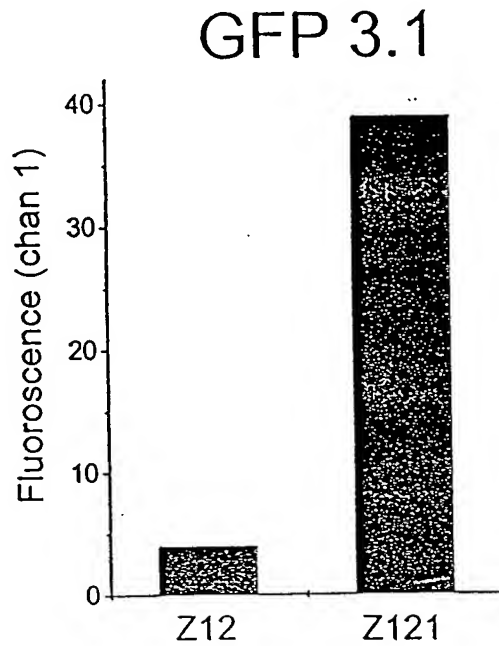
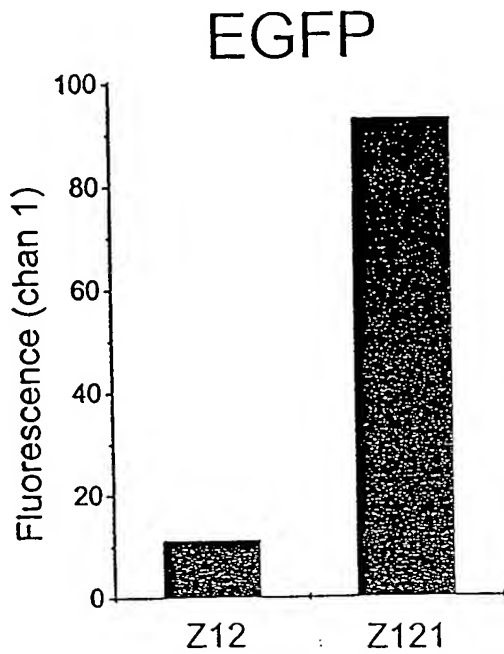


Figure 5.

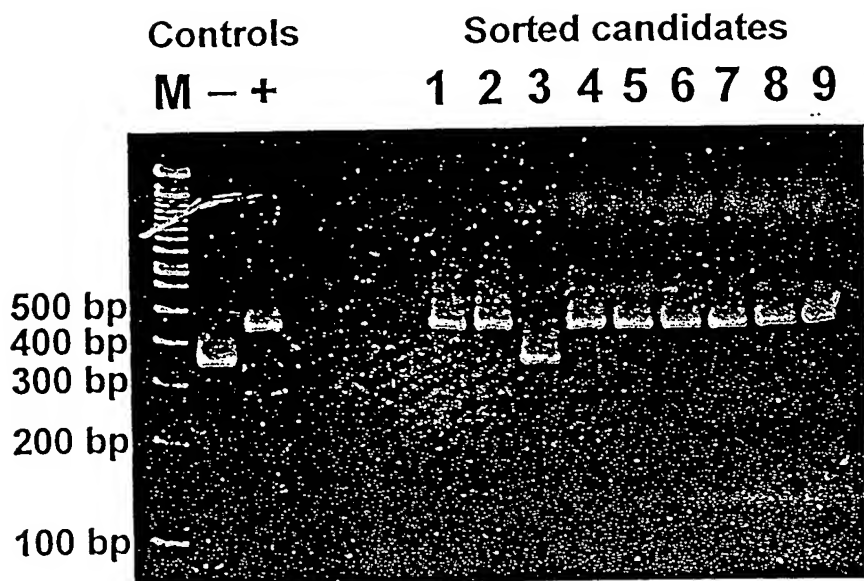


Figure 6

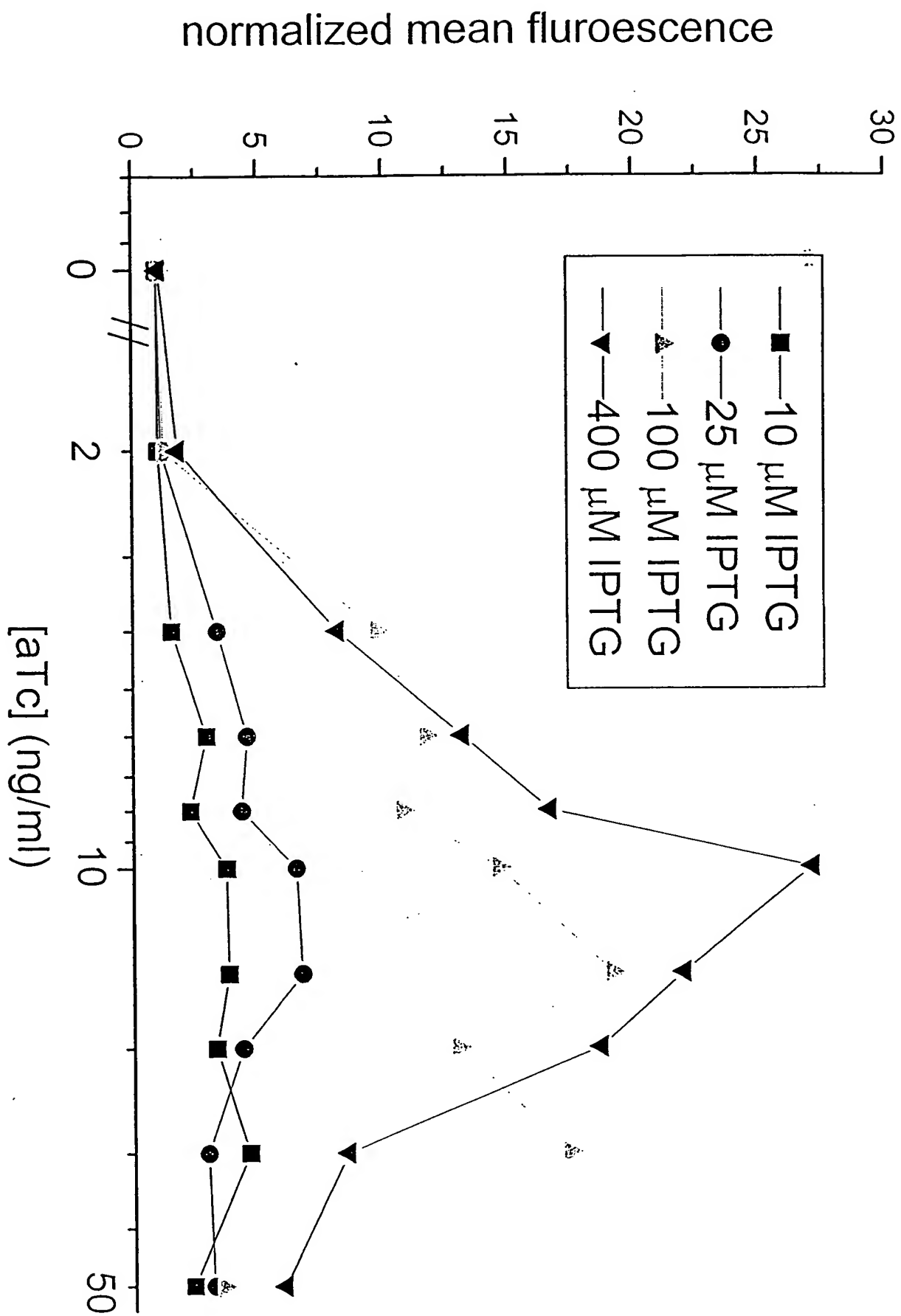


Figure 7

reporter construct #2

reporter construct #1

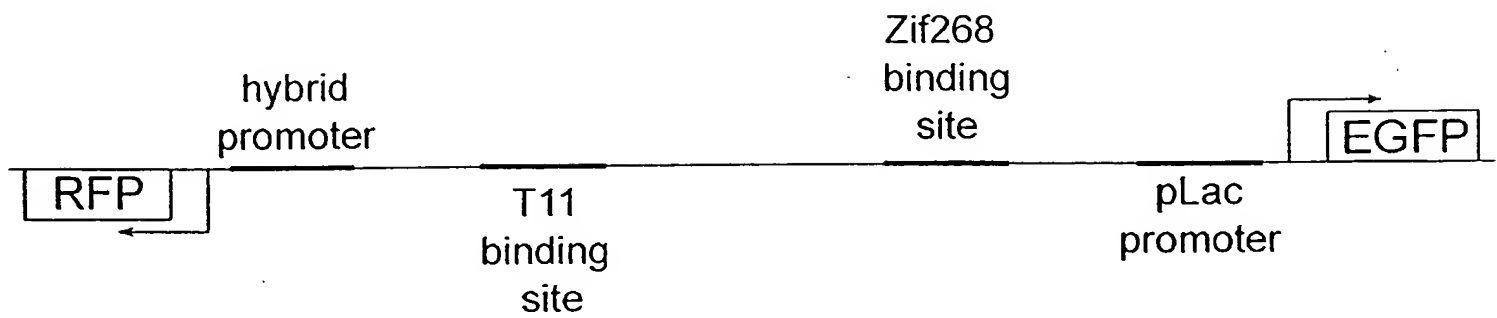
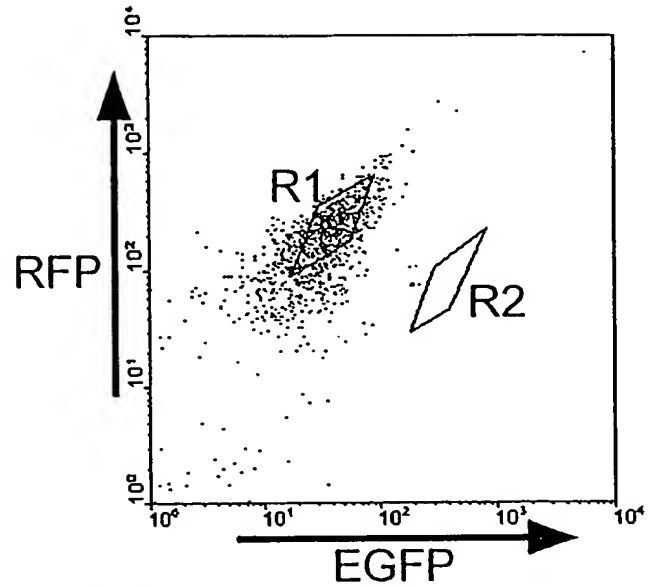
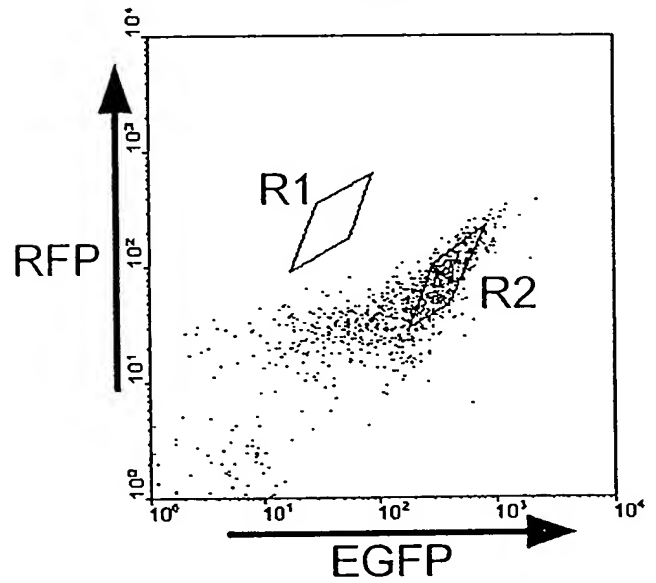


Figure 8

Two-Color reporter +
Gal11P-T11 protein



Two-Color reporter +
Gal11P-Zif268 protein



Two-Color reporter +
Gal11P-Z12 protein

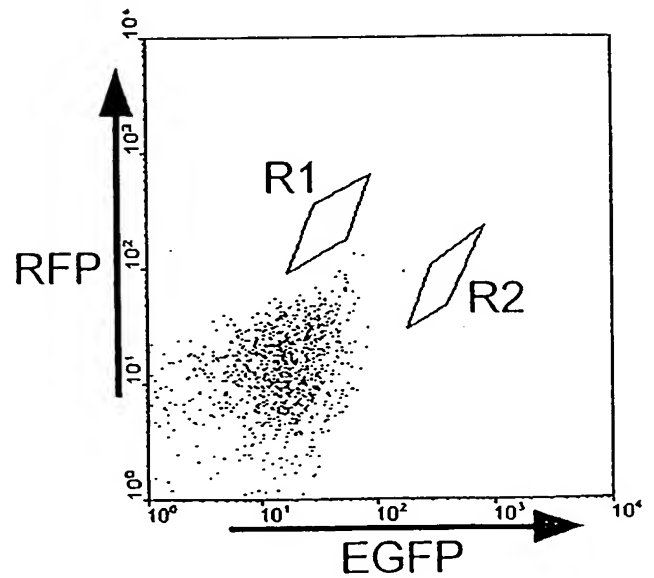


Fig 9

P53^{ZF} *in vitro* site selection consensus sequence:

CXGGACACGTX

(where X = no clear preference)

In vivo site selection library

CGGGANNNNNNG

(where N = a mixture of A, G, C, and T)

Selected Clones:

Sequence	# of clones
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CGGGAC <u>ACG</u> TG	9
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CGGGAC <u>ATG</u> TG	5
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CGGGAC <u>ACG</u> GG	2
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Sequence	Fold Activation
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CGGGAC <u>ACG</u> TG	18.6±2.7
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CGGGAC <u>ATG</u> TG	12.0±0.5
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CGGGAC <u>ACG</u> GG	12.6±1.9
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